Figure 2I



Supporting Information for Szyperski *et al.* (2002) *Proc. Natl. Acad. Sci. USA* **99** (12), 8009–8014. (10.1073/pnas.122224599).

Supporting Figure 10

**Fig. 10.** Experimental scheme for the 3D HCCH-TOCSY experiment. Rectangular 90° and 180° pulses are indicated by thin and thick vertical bars, respectively, and phases are indicated above the pulses. Where no rf phase is marked, the pulse is applied along *x*. The scaling factor k for $^1$H chemical shift evolution during $t_1$ is set to 1.0. The high power 90° pulse lengths were: 5.8 ms for $^1$H and 15.4 ms for $^{13}$C, and 38 ms for $^{15}$N. $^{13}$C decoupling during $t_1$($^1$H) is achieved using a ($90_x$ $180_y$ $90_x$) composite pulse. The lengths of the $^1$H spin-lock purge pulses are: first $SL_x$, 5.7 ms; second $SL_x$, 0.9 ms; $SL_y$, 4.3 ms. SEDUCE is used for decoupling of $^{13}$CO during $t_1$ and $t_2$ (rf field strength = 1 kHz), and GARP is employed for decoupling of $^{13}$C during acquisition (rf = 2.5 kHz). The $^1$H rf carrier is placed at the position of the solvent line at 0 ppm before the start of the first semiconstant-time $^1$H evolution period, and then switched to the water line at 4.78 ppm after the second 90° $^1$H pulse. The $^{13}C^a$ and $^{15}$N rf carriers are set to 38 and 120.9 ppm, respectively. The length of $^{13}$C spin-lock purge pulses denoted $SL_x$ are of 2 ms duration. $^{13}$C isotropic mixing is accomplished using the DIPSI-2 scheme (rf = 8.5 kHz). The duration and strengths of the pulsed z-field gradients (PFGs) are: G1 (100 ms, 16 G/cm); G2 (2 ms, 15 G/cm); G3 (300 ms, 8 G/cm); G4 (500 ms, 30 G/cm); G5 (100 ms, 16 G/cm). All PFG pulses are of rectangular shape. A recovery delay of at least 100 ms duration is inserted between a PFG pulse and an rf pulse. The delays are: $t_1$ = 850 ms, $t_2$ = 3.2 ms. $^1$H- frequency labeling in $t_1$ is achieved in a semiconstant-time fashion with $t_1^a$ (0) = 1.7 ms, $t_1^b$ (0) = 1 ms, $t_1^c$ (0) = 1.701 ms, $Dt_1^a$ = 33.3 ms, $Dt_1^b$ = 19.3 ms, and $Dt_1^c$ = -14 ms. $^{13}$C- frequency labeling in $t_2$ is achieved in a semiconstant-time fashion with $t_2^a$ (0) = 1120 ms, $t_2^b$ (0) = 62.5 ms, $t_2^c$ (0) = 995 ms, $Dt_2^a$ = 160 ms, $Dt_2^b$ = 125 ms, and $Dt_2^c$ = -35 ms. These delays ensure that a 90° first-order phase correction is obtained along $w_2$($^{13}$C). The fractional increases of the semiconstant-time period in $t_1$ equals to l = 1 + $Dt_2^c$ / $Dt_2^a$ = 0.58, and in $t_2$ equals to l = 1 + $Dt_2^c$ / $Dt_2^a$ = 0.78. Phase cycling: $f_1 = x$; $f_2 = x, -x$; $f_3 = x$; $f_4 = 2(x), 2(-x)$; $f_5$(receiver) = $x, -x$. Quadrature detection in $t_1$($^{13}$C/$^1$H) and $t_2$($^{13}$C) is accomplished by altering the phases $f_2$ and $f_3$, respectively, according to States-TPPI. For acquisition of central peaks derived from $^{13}$C steady state magnetization, a second data set with $f_1 = -x$ is collected. The sum and the difference of the two resulting data sets generate subspectra II and I, respectively, containing the central peaks and peak pairs.

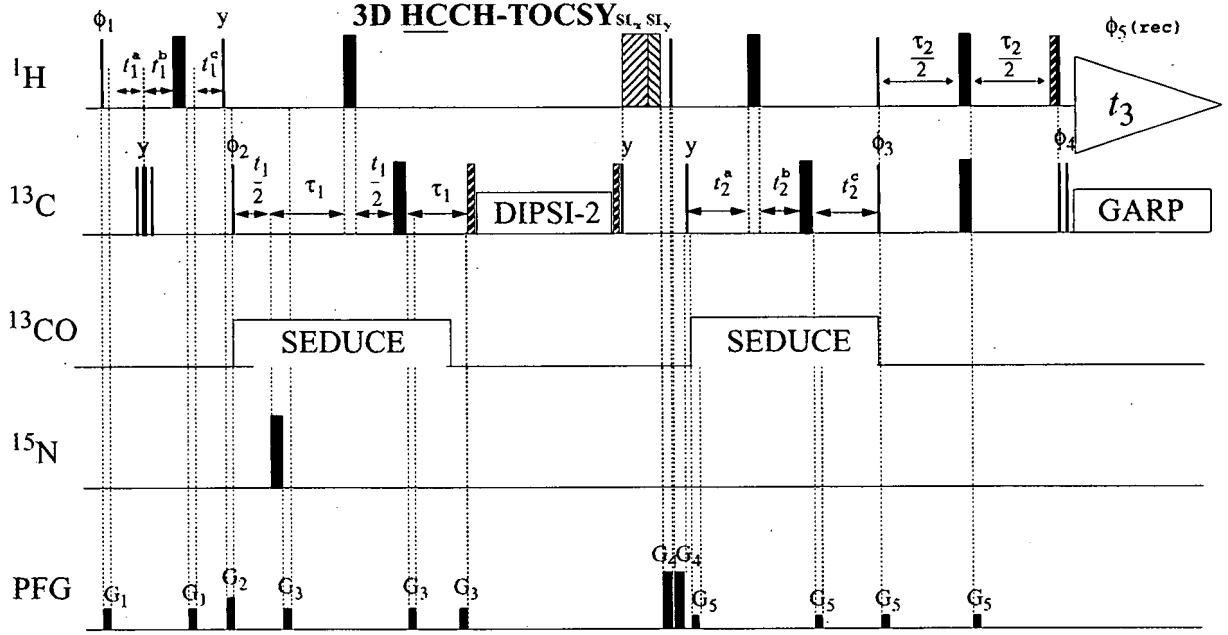
3D HCCH-TOCSY SL_x SL_y
ϕ1
y
ϕ5 (rec)
1H
13C
13CO
15N
PFG
DIPSI-2
GARP
SEDUCE
SEDUCE
ϕ2
ϕ3
ϕ4
τ1
τ2
t3
G1
G2
G3
G4
G5

Figure 10